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**GOVERNMENT OF SIKKIM
ANIMAL HUSBANDRY AND VETERINARY SERVICES DEPARTMENT
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No. 01/GoS/AH&VS/2022-23

Date: 5th April 2022

NOTIFICATION

Standard Operating Procedure on Control and Management of African Swine Fever (ASF) - To be Adopted by Field Veterinarians and Piggery Farm Managers.

Sikkim has recorded many cases of African Swine Fever (ASF) in various locations in Gangtok, Pakyong, Mangan and Namchi Districts by Real Time PCR at National Institute of High Security Animal Disease (NIHSAD), Bhopal vide letter no. F.No. 15-57/2012-NIHSAD, dated 29-03-2022.

ASF is a highly contagious and fatal disease of domestic and feral pigs (including wild boar) transmitted through direct and indirect contacts, ingestion of contaminated feedstuffs and by certain tick vector species. The disease does not infect humans (not Zoonotic) or other livestock species. However, no vaccine or drugs are available to prevent ASF infection at present.

Government of India directed that it is being an exotic and contagious disease to initiate all necessary actions towards rapid containment of the disease vide No: D.O No: K-11/22/2020-LH Dated: 1 April 22. In view of it the following Standard Operating Procedure shall henceforth be followed to control and manage African Swine Fever in the state. This SOP is in addition to Standard Operating Procedure (SOP) notified for PRRS vide No. 15/AH&VS/Adm/2021 Dated 7th March 2022.

African swine fever (ASF) is a highly contagious economically devastating hemorrhagic viral disease of pigs, warthogs and wild boar with high rates of morbidity and mortality. First described in the 1920s in Kenya, ASF is listed by the World Organization for Animal Health (OIE) as a notifiable disease. OIE reported first case of ASF in India on May 21, 2020 in Arunachal Pradesh and Assam. Later, the viral disease had spread to Mizoram, Meghalaya, Manipur and Nagaland, now in Sikkim. ASF does not pose a risk to public health (There is No zoonotic implications).

VIRUS CHARACTERISTICS: ASFV is an enveloped virus, 200 nm in diameter with a linear, double-stranded DNA genome that encodes 160 to 175 genes, belonging to Family: *Asfarviridae*. Genus: *Asfivirus*. It has distinctive morphology, characterized by the dense 80 nm virion core that is composed of the viral genome and an icosahedral capsid, covered by an internal lipoprotein envelope. The outer envelope is derived through the budding process from the cellular membrane of infected cells.

ASF virus is highly stable and temperature resistant and can persist in the environment for a long time.

Parameters	Resistance
Temperature	Highly resistant to low temperatures. Heat inactivated by 56°C/70 minutes; 60°C/20 minutes.
Meat and meat product storage	In the infected meat for several months at 4°C, in skin fat for 300 days, in salted, dried meat for up to 120 days, in ham in brine up to 180 days and in deep frozen (-70°C) meat/carcasses for several years and at -20°C for 2 years.
pH	Inactivated by pH 11.5 in serum-free medium. Serum increases the resistance of the virus, e.g. at pH 13.4-the virus survives up to 21 hours without serum, and 7 days with serum-supplemented media.
Chemicals/disinfectants	Susceptible to ether and chloroform. Inactivated by 8/1000 sodium hydroxide (30 minutes), hypochlorites-2.3% chlorine (30 minutes), 3/1000 formalin (30 minutes), 3% ortho-phenylphenol (30 minutes) and iodine compounds.
Survival	Extremely resistant to high temperature, putrefaction and desiccation. Virus remains viable for long periods in blood, faeces and tissues; especially infected, uncooked or undercooked pork products. Virus can also multiply in vectors; soft ticks of the genus <i>Ornithodoros</i> sp.

Susceptible Species: Pigs of all age group are susceptible to the disease, including all members of the pig family (Suidae): Domesticated swine (*Sus domestica*) and Wild boars (*Sus scrofa scrofa*). Warthogs (*Phacochoerus spp.*) and Bush pigs (*Potamochoerus porcus*). The virus survive in the pig population by following two types of cycles namely 'Domestic cycle' (between domestic pigs and *Ornithodoros* ticks) and 'Sylvatic cycle' (between warthogs and *Ornithodoros* ticks). The warthogs, Bush pigs and *Ornithodoros* ticks are considered to be reservoir of the virus.

RISK FACTORS ASSESSMENT: There are several risk factors which may affect the pig industry, livelihoods, nutritional security, socio-economic status and behavioural change of the farmers and export and import of pork and pork products. Some risk factors have been identified in context to our country which might serve as a connection for further emergence of ASF. The identified risk factors of ASF are determined keeping in view the social and ecological understanding of agriculture and food security of the India which depends on agriculture and livestock for their livelihood. Risk factors associated with African swine fever are Pig farming practice, Population density, Modes of transmission, Waste disposal, No treatment or vaccine, Movement of animals, Carcass disposal, Long persistence of virus, Breach in biosecurity, Trades, Wild pig contact and Soft ticks vectors

TRANSMISSION:

DIRECT: The virus may spread through direct contact via the oro-nasal route after contact with excretions from infected pigs, through ingestion of pork or other contaminated products. This transmission route requires the existence of large, continuous populations of pigs for the virus to remain in circulation. The virus is shed in saliva, tears, nasal secretions, urine, faeces, and secretions from the genital tract. Blood, in particular, contains large amounts of virus. The introduction of new pigs into a herd or piggery often results in individuals fighting and biting each other. In the case of free-ranging or scavenging animals, infection can result from contact with infected roaming pigs, wild boar (sylvatic cycle), their carcasses, or food leftovers. Additionally, using the same needle to vaccinate or treat several pigs can transmit the virus.

INDIRECT: Indirect transmission through contaminated feed and fomites. The virus is transmitted from one farm to the next almost exclusively due to human intervention, e.g. movement of animals or equipment, the feeding of infected materials, etc. Pigs can therefore become infected by contact with many different infected sources, mainly infected pigs, pork, and other pig-derived products (e.g. swill), and fomites (e.g. bedding). These infected animals and contaminated materials can be transported over long distances by vehicles and people.

VECTOR-BORNE TRANSMISSION: through soft tick (Ornithodoros ticks) in tick-pig cycle.

INCUBATION PERIOD: The incubation period represents the time from infection (i.e. when the virus enters the animal) to disease (i.e. when the animal shows clinical signs). It varies with the route of transmission, for direct contact with ASFV- infected pigs, the incubation period is between 5-12 days and when bitten by an Ornithodoros tick, the period can be less than 5 days. Virus excretion can begin up to 2 days prior to the appearance of clinical signs.

CLINICAL SIGNS:

Forms	Clinical Signs
Acute Form	Fever (42!), anorexia, listlessness, cyanosis, incoordination, increased pulse and respiratory rate, leukopenia and thrombocytopenia (at 48-72 hours), vomiting, diarrhea, and abortion in pregnant sows. - Any survivors become carriers for life.
Sub-Acute Form	-Slight fever, reduced appetite, and depression. -Abortion in pregnant sows is also possible and can be an early sign.
Chronic Form	-Weight loss, irregular temperature spikes, respiratory symptoms, necrosis of skin, chronic skin ulcer, arthritis, pericarditis and swelling of the joints. Pigs with chronic ASF will experience recurring episodes of acute disease, which could eventually lead to death.

PREVENTION AND CONTROL: As of today, there is no cure and treatment for the disease. There is no successful vaccine against ASF in the world. So, prevention is the only option to control the disease or limit its transmission. In the present situation prevention by adopting the strict bio-security guidelines is the only solution for this disease. Effective bio-security requires mostly change in the mind set, and therefore the changing behaviors of all peoples to adopt bio-security should be in partnership mode. Changing mindset starts by building a good understanding of the risk factors for ASF transmission.

BIOSECURITY GUIDELINES TO PREVENT INTRODUCTION OF ASF IN THE PIG FARM OR DISEASE-FREE AREA

- The disease free zone team members cannot travel to the Infected zone (IZ) at all.
- Segregation of the animals from the potentially infected animals and contaminated materials should be maintained and hence a proper fencing must be required surrounding the pig farm complex to prevent the access of wild boars and other wild animals. Double fencing of the periphery of the farm complex is recommended.
- The visitors and vehicles should not enter into the farm premises. If there is necessity then the vehicle's tyres & wheels and visitors' (any outside person) shoes/boots should be disinfected with 2 % sodium hypochlorite or 3:1000 Potassium permanganate solutions.
- There should be provision of foot dip at the main gate of the farm and also at entrance and exits of individual pig shed/pen. These foot dips with proper disinfectant solutions should be made mandatory at the entry point of each pig shed. There are several chemicals which can be used in foot dip like Potassium permanganate (3:1000), 2% Sodium hydroxide, 1% formaldehyde, 1% bleaching powder or 1% Calcium hydroxide (lime) etc
- Remove all moveable objects like utensils, tools, and clean and disinfects them separately, mechanically remove all visible dirts and organic matters by sweeping with a stiff brush (dry cleaning work) and then use water and detergents to moisten all surfaces (for some time period) and then rinse with high pressure water till no visible dirts should remain. Care should be taken to clean all corners of the pig sheds.
- Disinfect the pig shed and premises by ASFV approved disinfectants like 8:1000 NaOH (30 min), 1-2% sodium hypochlorite (30 min), 3:1000 formalin (30 min), 3% ortho-phenylphenol (30 min), 2-3% iodine compounds, quaternary ammonium compounds and some multiconstituent compounds (Virkon, Lysofromin, Verocid, Neogen® Viroxide Super) etc.
- The farm workers should properly wash their hands before entry into the farm and also after completion of work. The hands should be washed with any detergent based soap.
- The farm workers/personnel should wear specific dresses before entry to the farm till the completion of work.
- The farm workers/personnel and visitors leaving the farm after completion of work should ensure that their shoes, clothing and equipment are disinfected properly.
- The farm workers after completion of their work should wash their gumboots properly and disinfects theirs special dresses worn during work and keep their gumboots and dresses in a separate dry place (preferably under sun light as sunlight is a natural disinfectant).
- Separate the diseased animals from the healthy animals immediately.
- There should always be separate workers for handling of diseased and healthy animals.
- Pigs should be purchased only from authorized source. Do not purchase pigs from unauthorized or unknown source.
- The newly purchased pigs should not be mixed immediately with other pigs. The newly purchased pig should be kept in quarantine for 30-45 days and monitor their health daily for any observable sign of illness.

- After quarantine period pigs should be screened for the presence of disease and if found negative then only introduce into the farm
- Pigs should not be allowed to comingle with other pigs, wild boar and other animals.
- No diseased pigs should be sold to traders/ butchers.
- Meat produced from pigs originating from the surveillance zone (regardless of where they were slaughtered) is termed "Restricted Meat". Such meat receives a special mark (a crossed through oval health mark) and CAN NOT be sold fresh. It must be treated at a designated treatment centre and prior to treatment only handled at designated premises.
- Animals unsold at live animal market should not be returned to the farm premises.
- There shall be no movement of pigs, genetic material, meat, feeds from the Infected and Surveillance Zones into the Disease-Free Zone but the movement of pigs, genetic material, meat, feeds within the Disease-Free Zone is allowed.
- Swill feeding (kitchen waste, hotel waste, airport/seaport waste or meat meal) should be prohibited and if practiced, it should be boiled properly (>30 min) before feeding.
- Sharing of equipment among farms/villages should be discouraged without proper cleaning and disinfection.
- Farm utensils used for feeding of diseased pigs should not be used for healthy pigs. Movement of animals from one farm to another farm or from one village to another village or any animal fairs should be strictly prohibited.
- Loading and unloading should be take place outside the farm perimeter
- Movement of breeding boar from one farm to another farm or village should be strictly prohibited
- Practice of AI should be encouraged provided the liquid semen should be procured from certified farm or organization.
- Movement of personnel (veterinarians and farm workers) from one farm premises to another should strictly is prohibited. No visitor should be allowed to go inside the farms.
- Management of animal health including regular deworming and minerals and vitamins supplements.
- Regular spraying of acaricides to control tick is mandatory. There should not be any cracks and crevices in the farm buildings.
- Stringent environment friendly measures should be adopted for proper disposal of farm waste.
- Dead pigs, effluents and discarded parts from slaughtered pigs should be disposed of appropriately, out of the reach of wild or free-ranging pigs.
- Proper disposal of dead carcass with deep burial of 6 feet deep using lime/bleaching powder.
- Proper record keeping of animal health and entry and exit of visitors should be maintained.
- If any pig suffers from disease, it should immediately be separated from the healthy stock and should be reared in quarantine shed till it is fully recovered.

- Any suspected cases of ASF should be immediately reported to Veterinary doctors and to the District Veterinary Officer.
- Personal motivation to follow the bio-security guidelines must be given to the farmers and their initiative must be encouraged.

STANDARD OPERATING PROCEDURE FOR BACKYARD PIGGERY FARMERS

Apart from the above mentioned guidelines, the backyard pig farmers must follow the points listed below;

- There should be complete ban on the movement of live pigs, slaughtered pig and meat products inside and outside of the infected villages.
- There should be one house/pig shed outside of the village premises to keep the newly purchased pigs for at least 25-30 days. This may be a community type pig shed and after removal of pigs (after 25-30 days) it should be properly cleaned and disinfected
- All the pigs should be kept in close confinement. Do not allow pigs for scavenging.
- The floors of the pig sheds must be made up of concrete and cement. It is easy to clean the Pakka floor.
- Feed should be kept inside the house in a cool and dry place.
- Fresh roughage or feed harvested in areas with infected wild boars should be strictly prohibited
- Harvesting of bedding materials from the areas with infected wild boars is strictly prohibited.
- Kitchen waste should not be given to the pigs and if it is practiced, it should be properly boiled and it should keep for long duration.
- The kitchen waste should not be dispose off in the ways that may accessed by wild boars.
- Exchange of the boar for breeding purpose should be strictly prohibited.

STANDARD OPERATING PROCEDURE IN THE ASF INFECTED AREAS

There are two main objectives in the infected area are to prevent further spread of infection through quarantine and control of livestock movements and to remove the source of infection as early as possible through slaughter/culling of infected pigs, safe disposal of carcasses and decontamination.

- All the pigs within 1km radius (As per the National Action Plan for control, containment and eradication of ASF, DADF, GOI) must be slaughtered by humane methods, whether or not they currently show signs of the disease. Culling is undertaken by qualified persons, contracted by State/District veterinary authorities, under the supervision of a registered veterinary practitioner.
- The carcasses of dead/culled animals must be disposed off in the infected premises itself. The carcasses shall not come out from the infected premises.
- All the pigs should be immediately quarantined with complete ban on the exit of live pigs, pig meat and other potentially contaminated materials.

- The whole assigned team will only work on the Infected Zone and should not travel to the Surveillance Zone (SZ) or the disease free zone.
- Transport vehicles and other equipments must be disinfected before leaving from the infected areas.
- The area covered under 1 km radius from infected premises must be in the intensive active surveillance and frequent clinical examination of the pig herd must be carried by veterinary officers.
- It is the duty of Government officials that any smuggling of either live pigs or pig meat should not be practiced out of the infected areas and the trade of pork meat (fresh and products) within the "infected zone" is prohibited. The entry of pork meat (fresh and products) to the "infected zone" is only allowed from the "free zone". The exit of pork meat (fresh and products) out of the "infected zone" is prohibited.
- All the dead and slaughtered pigs must be disposed only by deep burial/incineration methods to ensure no leakage of the virus in the environment as per the FAO manual.
- Thorough cleaning and disinfection (2% Sodium Hydroxide, detergents, Sodium or calcium hypochlorite and iodine compounds) of the infected areas where animals were kept it includes-pig sheds, feed and water-troughs.
- The potentially contaminated materials like manure, bedding, straw and feedstuffs should be removed and disposed of in the same way as carcass.
- Semen, ova and embryos collected from pigs at the IP during the period between the probable introduction of ASF at the premises and the implementation of official measures will be traced and destroyed under official supervision in such a way so as to avoid the risk of spread of the African Swine Fever virus (ASFV).
- Do not allow pigs for scavenging system of feedings. Always keep pigs inside the close confinement.
- Wild boar movements and hunting should be prohibited in the infected areas and domestic pig should not be allowed to go inside forest areas.
- Safe disposal or removal of the infected wild boars carcasses from the forest area should be done and proper cleaning and disinfection of that particular area should be carried out immediately.
- After slaughter, disposal and decontamination procedures must be completed and premises left destocked for a period of minimum 40 days as per the OIE.
- After 40 days of the destocking period, if farmers wants to reintroduce the pigs than only 10% of the normal stocking rate on the previously infected farms.
- These reintroduced pigs must be observed closely for 6 weeks to ensure they are free from ASF, and then reintroduce the full population of pigs.
- Public-awareness and education campaigns should be organized (preferably in local language) in the infected, surveillance and also in disease free areas or zones.
- Compensation against culling of the animals must be given to the farmers without delay. It may help in the early reporting of the disease
- The ASF outbreak should be immediately reported to Veterinary doctors and to the District Veterinary Officer and samples should be collected by standard procedure and send to referral laboratories.

STANDARD OPERATING PROCEDURE FOR FIELD VETERINARY OFFICERS TO DEAL WITH ASF OUTBREAK

Apart from the above mentioned operating procedures/advisory, the field veterinarian should follow the following guidelines;

- Before entering in to the ASF suspected/affected farms or area, the veterinarian along with his team should wear personal protective equipment like apron, gloves, facemask and gumboots.
- The veterinary doctors along with his team must not go from infected zone to the surveillance zone and team assigned in surveillance zone must not go to the infected zone.
- The surveillance zone team may travel to the disease free zone under special circumstances (e.g. reassess the zone limits of the Surveillance zone compared to the free zone) only but must observe proper downtime of at least 24 hours.
- Undertake clinical inspection of each farm subunit, clinical examination of selected animals and necropsy of dead (or euthanized) animals. When conducting a clinical examination of suspected animals, systematic examination should be done.
- It is also important to note down the history, source of purchase of animals, source of feed, type of breeding and housing system, any clinical signs of illness, treatment provided if any along with vaccination schedule.
- Samples should be collected with proper care and send to referral laboratory as soon as possible with proper labelling, date of collection and history of the farm/animals.
- The Veterinary Officer should not open the carcass for Post Mortem examination as the virus spreads through blood and other discharges.
- The samples from sick animals in infected area will not be collected; however, the samples from new cases away from infected area can be collected for lab diagnosis.
- The Veterinary officers should organize awareness programme among the pig farmers and producers at regular intervals to make them aware about the alarming situation of the ASF, common signs and preventive measures of the disease.
- It is the duty of the state veterinary department to keep a strict vigil on the entry of any live pigs and their product from the neighbouring states most particularly the states that share borders with Myanmar, Nepal, Bhutan and China.
- Constructing a timeline is a useful way of representing the times during which infection and transmission of disease might have taken place, and therefore guiding an outbreak investigation. Timelines are used to determine time windows for introduction of the virus (based on the incubation period) and for spread to other premises (using the period of virus excretion).
- Once a timeline has been established, the next step is to use it for source and spread tracing in order to establish contacts that could have led to virus transmission during the calculated timeframe.
- Once possible sources of infection have been identified, it is important to prioritize them in order to carry out further epidemiological enquiries. This allows for rapid investigation and control of any contact liable to spread disease further.

- Veterinarian involved in outbreak investigations on a potentially infected farm should not visit another farm for at least 72 hours to prevent further spread of the disease.
- If there are no pigs on your premises the investigator may return home, shower, and thoroughly wash hair. All clothes worn that day should be soaked in disinfectant for 30 minutes and washed with water at 60°C. If there are pigs on your premises, complete this step elsewhere.
- While investigating an outbreak of ASF the Vet should enquire the neighbouring farmers or those who have purchased or sold animals recently, to ensure the risk factor of ASF transmission. Service providers who have visited the farm recently should also be notified.
- Besides interviewing the farmer, veterinarian should make a careful survey of the premises. The outer perimeter should be walked in order to establish any contact with neighbouring pigs or wild suids. It is often helpful to make a sketch map of the area, showing the location of animal housing, animal groups, entry and exit points and boundaries.
- Surveillance of the ASF has to be carried out along with pig farmers, pig producers. Regular reporting and dissemination of information should be done. All pig producers, especially in the areas surrounding infected foci, should be visited at least twice, with a two week interval, to ensure that no untoward deaths have occurred.
- The Animal husbandry staff should be thoroughly trained in their roles, duties and responsibilities in an ASF emergency and preparedness. More intensive training will need to be given to those who will be in key positions.

POST-MORTEM FINDINGS: As ASF is classified as haemorrhagic febrile disease of viral origin, therefore, lesions or haemorrhages occur throughout the body such as on the tip of the ears, hind legs and quarters of the body. After opening of the carcass haemorrhages can be find out on lymph nodes, kidneys, larynx, bladder, colon, and gall bladder. Spleen may be enlarged. Excess pericardial and other body cavities fluid.

LABORATORY SAMPLES, DIAPATCH AND TRANSPORTATION

Sample	Purpose	Storage and shipment procedure
Whole blood	Virus isolation, PCR and ELISA	<ul style="list-style-type: none"> -Blood should be collected from febrile pigs up to 5 days after onset of fever -Blood should be collected aseptically from heart into EDTA/Heparin tubes -Blood should be stored at refrigeration temperature (4°C)
Lymph nodes, spleen, tonsils, lungs, heart and kidney	Virus isolation, PCR, ELISA	-Collected aseptically and kept chilled into ice but not in frozen condition.
	Histopathological Examination	-Collected in 10% Buffered formalin for histopathological examination and detection of virus by immunoperoxidase test.
Serum	Antibody detection	-Collected in a sterile vial after mild ³ centrifugation and kept in frozen condition (-20°C or -80°C) and transported in ice pack with proper labelling (Place, Animal No., Age, Sex)

Note:

- Samples should be stored in leakproof, water-resistant, sterile containers.
- All the samples should be transported into thermally insulated boxes with frozen ice packs or frozen gel packs.
- The samples are transported as per the condition mentioned in the table above; as early as possible to referral disease diagnosis laboratory for ASF (Regional Disease Diagnostic Laboratory, RDDL and National Institute of High Security Animal Diseases, Bhopal) with proper labelling like Name of owner, locality, brief history, animal No., date of collection, Age, Sex, Breed etc.

SOP FOR STAMPING OUT POLICY FOR ASF (INCASE OF CONFIRMED POSITIVE CASES):

Zoning: Infected premises (IP) means an epidemiological entity where pigs have become infected. It may be a single farm or household or entire village and settlement. It may be a livestock market or abattoir.

A dangerous-contact premises is one for which there are epidemiological ground to suspect that it has become infected even though the disease is not yet clinically apparent. This might be through closed proximity or as a result of epidemiological tracing.

ZONING OF INFECTED AND HIGH RISK AREAS:

Zone	Objectives	Action
Infected Zone (IP)	Prevent further spread of infection	<ul style="list-style-type: none">-Culling of all pigs within 10 kms of positive confirmed cases (OIE).-Quarantine and ban of livestock movement, exit of live pigs, pig meat and other potentially contaminated materials.-Pig markets and abattoirs to be closed
Surveillance Zone	Enhanced active surveillance	<ul style="list-style-type: none">-Pigs should be inspected at weekly interval and owners to be questioned about disease occurrence, pig movement, etc.-Movement of pigs, pig meat, pig products from IP should be banned.-Movement from surveillance to free zone may be allowed but only after health inspection and issue of permit.-Abattoir and pig meat processing plant may be allowed to operate but must be subjected to strictly enforce zoosanitary codes of practices.-Sale of live pigs and pig meat may be allowed to continue unless they constitute a treat of further spread of the disease.
Disease Free Zone	Preventing entry of the disease.	<ul style="list-style-type: none">-Entry of pigs and pig products from infected zone should be banned. (Allowed only subjected to official permit from surveillance zone.)

Controlling the disease is a shared interest and must be considered a shared responsibility. Veterinary services, which include public and private sectors in both affected and unaffected countries, have the mandate to safeguard animal health and welfare and should lead the implementation of effective coordinated countermeasures to minimize the economic loss due to the disease.

BY ORDER

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